

The University of Chicago

THE HISTOLOGICAL ANATOMY OF
THE HYPOCOTYL OF CUCUR-
BITA MAXIMA DUCHESNE

A PART OF A DISSERTATION SUBMITTED
TO THE GRADUATE FACULTY IN CANDIDACY
FOR THE DEGREE OF DOCTOR OF PHILOSOPHY

DEPARTMENT OF BOTANY
1930

By
RAY WATSON RUTLEDGE

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INTRODUCTION

There has been much disagreement in attempts to interpret the nature of the bundles in plants that have an internal phloem, as to whether they are or are not bicollateral. Many of the investigations on the anatomy of the Cucurbitaceae have been made in attempts to determine the ontogeny and phylogeny of the internal phloem, but very few of them deal with details of the cellular structure of the plant as a whole or any considerable part of it.

Hérail (4), as a result of his studies on the comparative anatomy of the stem of many dicotyledons, concluded that the Cucurbitaceae alone have bicollateral bundles.

Baranetzky (1) investigated the ontogeny and later development of the bundles of the stem of several members of the Cucurbitaceae, and concluded that the internal phloem represented separate incomplete bundles with a unilateral cambium. He also stated that this cambium may give rise to xylem, forming a complete internal bundle.

Von Faber (9), on the basis of his studies on the development of the bicollateral bundle at the apex of the stem of *Cucurbita pepo*, reported that the external and the internal cambiums appear in the bundles before any xylem elements are visible.

Fischer (2), in a rather extensive study of the

phloem system in this family, distinguished four types of phloem: (1) hypodermal or ectocyclic, between the epidermis and the stele; (2) fascicular phloem; (3) entocyclic, within the stele; and (4) commissural phloem, which connects the other types of phloem. He stated that in the transition the internal phloem ends blindly at its lower extremity.

Lamounette (6) has studied the ontogeny of the internal phloem in relation to the procambial strands in the hypocotyl, cotyledons, leaves, and stem of various members of the Cucurbitaceae. He reported that the internal phloem in the stem was not derived from the same procambial strand as the other tissues of the bundle, but from a secondary procambial strand formed internally to the primary strand and slightly later. He also found that, in the transition region of *Cucurbita maxima*, there was a fairly large amount of external phloem differentiated before any internal phloem was formed. He concluded that the internal phloem was an abnormal development resulting from the activity of the pith cells.

More recently Worsdell (11) investigated the medullary phloem in the Cucurbitaceae from a phylogenetic viewpoint, limiting his work almost entirely to the axis after secondary thickening had begun. He regarded the medullary phloem as a vestigial structure, "the remnant of a former system of medullary vascular bundles in which the xylem has disappeared."

Of the more general treatises on the morphology and

anatomy of various members of the Cucurbitaceae, the root has been described by Van Tieghem (8), the transition from root to stem in *Cucurbita maxima* by Gérard (3), and the course of the bundles in the stem by Weiss (10) who concluded that all the bundles in the stem are leaf traces. More recently Manteuffel (7) followed the course of the bundles in *Cucurbita pepo*, and stated that the five bundles of the inner ring are cauline, and that the five bundles of the outer ring are common bundles. Yasuda (12) has compared the general anatomy of the Cucurbitaceae of Japan with one another, but no detailed consideration of the stelar anatomy was given.

Many of the members of this family have been described by Zimmerman (13, 14), but his work was chiefly on the physiology of the axis. Holroyd (5) has investigated the transition region and, more briefly, the secondary structures in *Cucurbita pepo*, as well as some other members of the family.

Although a large amount of work has been done on the anatomy of various members of the Cucurbitaceae, the general viewpoint has been mainly phylogenetic. But little effort has been given to a thorough histological study of the primary tissues of the transition region, of the structures involved in secondary thickening, or of the tissues derived from such activity. The present paper is an attempt to furnish information on these points.

I. THE PRIMARY BODY

Most of the work on the development and anatomy of the primary structures is based on a study of the young plant at the stage where the hypocotyl has attained a length of about 10 to 15 millimeters. This is a fairly young stage since the hypocotyl, when fully elongated, may attain a length four to five times that of this stage. But it is at this fairly early period that, at most levels, the cambium has begun to develop. Younger stages were used to determine the relative time and order of development of the tissues. The youngest used was the hypocotyl about 1 to 2 millimeters long, just emerging from the seed coat. At the 10 to 15 millimeter stage the primary root has developed considerably in length, with many secondary roots. But all the tissues of the root, particularly the xylem, have not completely matured at this time, so for the mature primary tissues of the root a slightly older stage was used. When the hypocotyl is 15 millimeters long the upper part is just straightening out, or it may be still recurved. The cotyledons have not expanded. The peg is well developed. The entire hypocotyl down to the peg is covered with hairs, both glandular and spinulose, with perhaps more of the former. The hypocotyl is somewhat flattened in the plane parallel to the cotyledons so that a cross section presents an oval outline. The peg is formed on one or both sides of this flattened axis.

The stele of the primary root is a tetrarch, exarch protostele. The xylem is composed of elongated conducting

elements and parenchyma that becomes reticulate-pitted. In the middle of the cylinder there are two, three, or four (usually two) large reticulate-pitted tracheae, surrounded by the smaller, scalariform, reticulate or pitted thickened parenchyma cells. These cells are almost isodiametric, or slightly elongated transversely. The metaxylem immediately within the protoxylem is composed of long, slender, scalariform, and scalariform-reticulate tracheae. The protoxylem is composed of very long, very slender spiral and annular elements that are crushed early in most cases, though the cells may be seen in much later stages.

The primary phloem of the root is arranged in four groups radial to the protoxylem points. The protophloem is composed of parenchymatous cells, most of which become elongated, and some of which become very long. No sieve tubes are found in the protophloem. The band of protophloem is only one, two, or three cells wide in most cases. The cells themselves have a very small diameter. The metaphloem constitutes the larger portion of the primary phloem, and is composed of sieve tubes, companion cells, and parenchyma.

The pericycle in the root comprises a layer of parenchyma one cell thick over the phloem groups, and usually four to six cells thick over the protoxylem points. The cells are mostly very long vertically, having a length several times the radial diameter.

The endodermis is a single layer of cells just external to the pericycle. The cells are quite long vertically, being about the same length as the cells of the

pericycle. The Casparian strips are narrow, but distinct even in rather young roots. The remainder of the cortex is a layer of parenchyma about seven to ten cells in thickness in most cases. The cells have a fairly large radial diameter, by far the largest in the root, except the large tracheae at the center. The intercellular spaces are conspicuous (as seen in cross section). Vertically the cells have a length several times as great as the radial diameter. No pits were observed in the walls. The epidermis is a single layer of cells, small in radial diameter, but rather long vertically. In the region of the primary root, just below the transition, no hairs of any sort were seen.

The root-stem transition includes all the axis between the first change in the structure of the root in the direction of stem characters upward to the point in the axis where the stem characters are definitely established. In this plant, the latter is the region where the endarch condition in the xylem is attained. The transition involves about one centimeter of the axis, roughly from the upper part of the peg to a level 11 or 12 millimeters below.

The first change from the root to the transition is noted about 11 to 12 millimeters below the peg. At this level the central tracheae extending into the root and two or more similar tracheae lie around a small pith of parenchymatous cells. The number of these large tracheae varies from four to seven, usually being four, five, or six. Two or three of these added tracheae may not mature until the hypocotyl has

completely elongated and some secondary thickening has been added. A pith of parenchymatous cells is present in the middle of the axis. These parenchyma cells are roughly circular in outline, as seen in cross section, with conspicuous intercellular spaces and with a vertical length two to ten times the radial diameter. Numerous small to rather large rounded and oval pits are scattered over the walls. Each large trachea is surrounded by small, almost isodiametric, reticulate-pitted thickened parenchyma cells such as are found in the root. In hypocotyls that are completely elongated these tracheids and tracheae, together with the smaller scalariform vessels just internal to the protoxylem may form an almost continuous ring of lignified xylem around the increasing pith. In hypocotyls about 15 millimeters long there is still considerable parenchyma between the tracheids.

At a slightly higher level these large central tracheae are in a position just internal to the cambium initials; the protoxylem points are farther apart, the entire stele is larger in diameter, and the pith is of rather large size. There is now more parenchyma between the large tracheae and the tracheae subtending the protoxylem. The endodermis is still distinct as a complete band, and the other tissues of the axis are practically unchanged, except that the phloem groups are larger in cross section.

Continuing up the axis the next important difference seen in its structure is the appearance of the internal phloem. This first appears at about 5 to 7 millimeters below the middle of the peg and about 4 to 6 millimeters above the

lower level of the transition. This level is about the middle of the linear extent of the transition. The internal phloem at this lowest point is composed of a few widely separated cells of parenchyma, very small in radial diameter, but considerably elongated vertically. Some of the cells have dense cytoplasm, while others have scant cytoplasm. They are very irregularly arranged in the central parenchyma, but usually lie in a rough circle three to four cells internal to the xylem. This is at about the level where the first spreading of the scalariform elements subtending the protoxylem is noted. The internal faces of the four original protoxylem groups are extended laterally in the direction of the large reticulate-pitted tracheae which lie just internal to the cambium initials. The remainder of the axis at this level is essentially unchanged from the last level noted.

At a point 1 to 2 millimeters farther up the axis the group of scalariform elements subtending each protoxylem point is divided definitely into two arms extending laterally in two fairly definite rows of cells. The position of the spiral and annular vessels is as yet unchanged, though there are in some cases two irregular rows of these elements, as contrasted with the one row found at lower levels. At this point, in addition, a few scattered, long, slender scalariform elements are present external to and between the large tracheae. There are now usually four pairs of these large tracheae, though there may be as many as four in a group, and they are much smaller than the central tracheae in the

root. At this same level, in most cases, the internal phloem forms a rather complete circle about two to three cells internal to the xylem. And these parenchymatous cells between the phloem and the xylem have, in many places, even in the 10 millimeter hypocotyl, begun to divide, forming an internal cambium.

Within a short vertical space after the two arms of scalariform vessels are formed, the group of spiral vessels also divides into two arms in line with the scalariform vessels. The two rows of spiral vessels may be formed a short distance below the point of splitting or they may be formed in the process of splitting. In either case, at a level about 1.5 to 2 millimeters below the middle of the peg, four definite pairs of groups of protoxylem-metaxylem elements are formed with the protoxylem elements still in contact. Almost immediately, however, the paired protoxylem-metaxylem strands separate, and the intervening space is occupied by parenchyma. At the same time more long scalariform elements are added between and internal to the large reticulate-pitted tracheae lying just internal to the cambium initials. There are at this level three to five of these large tracheae and they are much smaller than those in the root. The result of these developments is the rapid formation of four very nearly complete rows of lignified xylem elements, each with a protoxylem point at both lateral ends, and distinct from each other. At the same level the internal phloem is arranged in four fairly definite groups, each internal to a xylem group,

completing what might be termed the four transition bundles. The nature of the internal phloem at this point is in doubt. On its internal face there appears to be a rather irregular zone of parenchyma cells that may correspond to the external protophloem. But also, at a few points, sieve tubes were observed lying next to the pith. More work is needed on this point. Most of the internal phloem consists of sieve tubes, companion cells, and parenchyma and appears to be of the same nature as the metaphloem of the external primary group. The external phloem has not changed markedly except that the protophloem is not so distinct as in the root. A zone of cambium initials lies between the xylem and the external phloem, and there is also a more or less discontinuous zone of cambium initials appearing between the xylem and the internal phloem in some, but not all cases. Thus the transition bundles appear to be definitely bicollateral. In addition the external phloem has become extended into the pericycle over the point originally occupied by the protoxylem and a few more or less widely separated phloem cells are differentiated in these four rays and connect the internal and the external phloem. Sieve tubes are present in these connecting strands. Also, in one or two cases, a few scattered phloem parenchyma cells appear in the middle of the pith. The endodermis is still distinct as an uninterrupted band. The pith is much greater in diameter, and the entire axis has increased considerably in size.

The four transition bundles continue up the axis for

a very short distance (usually 0.75 to 2 mm.) before any important change occurs. About 0.75 to 2 mm. below the middle of the peg the first indication of the breaking up of the transition bundles is seen. At this level the axis is much larger, and the cortex is increasing rapidly in amount on one side as the peg is approached. The stele is larger and the transition bundles are farther apart. Phloem extends across the medullary rays between the bundles with connections to both the internal and external phloem. The endodermis is still distinct as a continuous band. The first indication of the breaking up of the transition bundles is the maturation of some of the cells in the middle of the xylem as parenchyma. An extremely short distance above this many more cells have matured as parenchyma, and the two xylem groups swing gradually away from each other, widening the gap. Within a very short distance the cells external to this new ray, in line with the cambium initials, mature as parenchyma and phloem, and within a vertical distance of 0.2 mm. some of the cells in the ray mature as phloem. Thus the external and internal phloems are connected on both sides of the new bundles (since the opposite side was connected some distance below). This process is the same for all the transition bundles, though there may be as much as 0.75 mm. distance vertically between the levels where the first and the last break up.

The ultimate result of the process just described is the formation of eight bicollateral bundles with phloem along the lateral faces connecting the external and the

internal phloems. The endodermis is still distinct as a continuous layer of cells. These bundles are not yet truly endarch, however. Though the protoxylem is internal to the large reticulate-pitted tracheae, it is at one side of the smaller scalariform metaxylem elements. The true endarch condition is attained gradually though a vertical distance of as much as one millimeter, though in some bundles within a much shorter distance. Usually the process is rather irregular and involves the gradual maturation of the protoxylem more and more toward the middle of the inner face of the bundle, and of the slender scalariform elements of the metaxylem more and more in the opposite direction toward the middle of the inner face of the bundle and external to the protoxylem. In a few cases the process is more regular, the protoxylem maturing through an almost perfect arc of 45 degrees, resulting in an endarch condition. The bundles become endarch in all respects at about the level of the upper part of the peg or slightly above it.

The peg is a local development of the cortex. It is developed either on one side of the flattened axis, or principally on one side and slightly on the other. Anatomically it consists of ordinary cortical parenchyma. The cells tend to be radially elongated, particularly in the lower part.

At this level there are, then, these eight bicollateral endarch bundles, fairly regularly spaced in a circle or oval. The bundles are usually about the same size, as seen in cross section, but there may be a little variation. In

the xylem there is considerable parenchyma separating the metaxylem elements, and, in many cases, the protoxylem elements from each other and from the metaxylem. The larger reticulate-pitted tracheae from the middle of the root are here represented by one, two, three or four small tracheae of a similar nature lying just internal to the cambium initials. The phloem is similar to the phloem at the level where the eight bundles were formed except that a small group of cells just internal to the protophloem matures as fibers. They do not, however, mature until a small amount of secondary thickening has been formed. The fibers are small but extremely long. There are many bordered pits in the walls. There is a large mass of external phloem and very little internal phloem. The internal cambium has practically disappeared. These bundles continue up through the hypocotyl practically unchanged in structure to the cotyledons, except for the branching and anastomosing that may occur, which will be discussed later. The endodermis can be determined with certainty only over the bundles, where the Casparian strips can be seen. Its position over the rays can usually be determined vaguely by the fact that some of its cells are filled with starch grains. The pericycle comprises the single layer of cells between the phloem and the endodermis. But at some points in some of the bundles the phloem adjoins the endodermis and the pericycle is lacking. The amount of interfascicular phloem varies widely, though in almost all cases it forms a fairly complete, close or rather loose network of

branches, as seen in tangential section. There are connecting strands with the external and internal fascicular phloems. The strands are composed of sieve tubes, companion cells, and parenchyma. The pith at this level is very large, occupying somewhat more than half the axis. The cells are circular or hexagonal in outline, with intercellular spaces, and rather elongated vertically. In some specimens there are phloem patches of two to four cells each widely distributed and apparently with no definite arrangement. The strands end blindly at their lower extremities and continue through the hypocotyl up to the cotyledonary node where it is impossible to trace them farther. There appear to be connecting phloem strands between them and the interfascicular phloem, though this was not definitely demonstrated.

The cortex just above the peg is about twelve cells in thickness. The cells are fairly large in cross section near the stele, circular or roughly hexagonal in outline with small intercellular spaces. But the outermost zone of about four cell layers is composed of much smaller cells more definitely circular or hexagonal, and more closely packed. Stomata are present from the peg to the cotyledons. The cortex is essentially similar in structure from the peg to the cotyledons. The cortical phloem is discussed later.

At about the level at which the bundles become truly endarch, or in some cases slightly below it at the peg, additional bundles may arise. At their lowest end these new bundles are composed entirely of phloem. Within a short

distance above this point (0.2 mm. or less) one or two spiral elements appear internal to this phloem strand. In some cases the xylem appears internal to all the phloem except the innermost cells, thus making the bundle bicollateral. The internal phloem, when present, consists of two or three parenchyma cells. Very slightly above this, other protoxylem elements are present, either lateral to or external to the first. And almost immediately above this a few metaxylem elements may be found, though they may not be present for a short distance up the axis (0.4 mm.). At the level at which the metaxylem elements appear there is always a small amount of internal phloem and a large amount of external phloem which seems to be similar in composition to the external phloem of the eight original bundles. The composition of the phloem at this level is in doubt, although it appears to be mainly of parenchyma. There are also phloem strands between the external and internal phloems around the lateral faces of the bundles, though these may be somewhat discontinuous. A zone of cambium initials develops between the external phloem and the xylem but there is no indication of an internal cambium at this early stage.

The number and distribution of these additional bundles are variable but it seems that usually there are one or two. Generally the hypocotyl above the peg is oval in outline and the eight main bundles are arranged roughly in two groups of four bundles each, each group more or less toward one end of the oval. Frequently it is between these two groups of bundles that the additional bundles are found,

often one on each side of the pith. However, they may be between almost any two of the eight bundles that extend to the transition. Thus a very short distance above the peg the number of bundles is usually increased to ten.

The total number of bundles is ordinarily still further increased by the branching of a variable number of the original eight bundles. In many cases two of these bundles branch, bringing the total number to twelve. However, the number of bundles just below the cotyledons varies from eight to fourteen in my preparations. The larger number usually results from the branching of the original bundles. The smallest number of bundles found in the upper part of the hypocotyl, viz., eight, is the result of a peculiar process of bundle "fusion". In this hypocotyl, at the upper part of the transition, eight bundles are formed as usual, quite distinct from each other. But almost immediately the pair of bundles at each end of the oval cross section of the hypocotyl merge, forming a large bundle at each end of the oval, and decreasing the total number to six. (These bundle pairs are derived from different "transition" bundles, but may be regarded as having been derived from the same protoxylem-metaxylem group in the root). These six bundles continue up the axis about 2.5 mm. above the peg, at which point one of the large fusion bundles branches to give one smaller one between two very small ones. Thus the number of bundles is increased to eight. Shortly above this the other large fusion bundle branches similarly increasing the total number to ten, two on each side of the

oval axis and three at each end, as seen in cross section. These ten bundles extend up the hypocotyl to a point a short distance below the cotyledons where the three bundles at each end of the oval merge to form a large bundle which, almost at once, branches to form two bundles. Thus the number of bundles just below the cotyledonary node is eight.

In most of the specimens examined cortical phloem was present. The lowest point at which it is found is at the peg. Here it is present as widely separated groups of cells of two to four cells each, on the same side of the axis as the greatest development of the peg. Some of the phloem strands are found extending out into the peg itself. These strands at this level appear to be formed of phloem parenchyma, the presence of sieve tubes not being definitely established. Shortly above this level the phloem strands are found extending farther around the axis, and at a slightly higher level they can be seen in the entire circle of the cortex. At this higher level (just above the peg) the strands may consist of two to five cells, as seen in cross section, the most common numbers being three and four. The cells are small in diameter, mostly rather long and slender, some with dense cytoplasm. The strands at this level seem to be composed mainly of sieve tubes and companion cells, with some parenchyma. They form a much-branched complex of anastomosed strands distributed from the stele to a region two or three cells within the epidermis. They end blindly at the peg and extend through the upper part of the hypocotyl to the cotyledonary node, finally

extending out into the cotyledons. The system has strands connecting it with the phloem in the foliar ray which is connected with the external and internal fascicular phloem, and possibly with the phloem in the pith, if any is present. Thus the cortical phloem completes a complex phloem system that is distributed through all parts of the hypocotyl from the peg to the cotyledons.

II. THE SECONDARY BODY

1. The Pith

The pith in the lower part of the transition region becomes active soon after secondary thickening begins. When the axis is only a little more than one millimeter in diameter many of the pith cells have already divided. This occurs even at the lowest level of the transition. A few small strands of phloem are formed as a result of these cell divisions.

As the axis increases in diameter the pith continues to be extremely active, becoming very large. This pushes the elements of the primary xylem widely apart, and with the accompanying activity of the parenchyma in the primary xylem, separates them in varying ways at various points. (In some cases the cells between the lignified primary xylem and fascicular cambium do not lignify and become active later, separating widely the primary and secondary xylem in the root and lower transition).

The tertiary phloem formed in the pith shows a wide variety of arrangements. In some cases there are a few very large definite phloem strands, seemingly formed in an eccentric manner with a layer of cambiform cells surrounding them. In other cases somewhat smaller strands are grouped variously and extend in many directions. Some have no perceptible cambium zone, while others have a cambium at one side with no specific orientation. These strands are branched, anastomosed,

and form a very complex network. In still other cases, especially at a higher level in the transition, most of the tertiary phloem is arranged in a large circle with a definite external cambium completely surrounding it, and at some distance from the primary xylem. In this last type there are also small phloem strands, some apparently primary, some obviously tertiary, within this circle of phloem, and also between it and the primary xylem. All this phloem is composed of sieve tubes, companion cells, and a fairly large amount of parenchyma. These phloem strands seem to end blindly at their lower ends and extend for varying distances up the axis, finally connecting with the transverse phloem strands in the foliar rays and with the internal phloem of the bundles.

A few of the older hypocotyls (two to three months old) have complete collateral bundles in the pith with xylem, phloem, and cambium. The xylem consists of one, two, or three large tracheae and fibers or parenchyma, or merely of a small strand of fibers. These bundles may be oriented in almost any direction. They end blindly at their lower extremities, and in most cases extend slightly above the transition. At the upper end the xylem and phloem may be connected with the external fascicular xylem and phloem respectively, by a collateral bundle extending laterally through the foliar ray.

In the upper part of the hypocotyl the pith cells increase in size enormously as the plant grows older, but

there are not large numbers of cell divisions. There are a few small strands of tertiary phloem present.

2. The External Cambium and Its Derivatives

At the time secondary thickening commences the cambium in the young root extends almost to the protoxylem points. Although the pericyclic cells over the protoxylem points soon become active, the cambium, as a definite zone, is not found in the broad xylem rays over the protoxylem points until a fairly large amount of secondary xylem has been formed. As the secondary bundles increase radially, the fascicular cambium is extended gradually laterally by the activity of the adjoining ray cells, which, by dividing regularly periclinally, form a definite cambium. Consequently the bundles are very wide externally and the rays are about the same width throughout.

The secondary xylem is composed of many tracheae, numerous fibers, parenchyma, and lignified parenchyma. The tracheae are extremely long and of very large diameter. They are composed of many short segments. The numerous half-bordered pits are oval or rounded, and arranged irregularly between prominent scalariform thickenings. Each trachea is surrounded by one to three (or more in some cases) rows of thickened, short, sometimes laterally elongated cells with many simple pits. These cells do not die as the xylem matures and some may later extend the closing membranes of the pits

into the tracheae, forming tyloses. These tyloses may completely block a trachea throughout its length. In old roots many of the tracheae may be so filled, especially those near the inner part of the bundle.

The fibers in the secondary xylem are very long, slender, with tapering ends. The walls have fairly numerous bordered pits with rounded, oval, or slit-like openings. The number and arrangement of the fibers vary widely. In some cases the xylem is composed largely of tracheae and fibers, particularly toward the inner part of the bundle, while in others the fibers are in small groups with no specific distribution. In any case each trachea is nearly always surrounded by a zone of fibers. The amount and distribution of the xylem parenchyma is also very variable. In the more nearly mature part of the old bundle there is usually very little parenchyma; in the outer region of the bundle there is usually much more. Some of the parenchyma comprises a few very definite xylem rays; some is irregularly distributed in groups between the tracheae. The cells of this parenchyma are mostly small, having a diameter slightly greater than that of the fibers. They are almost isodiametric or about twice as long as the radial diameter, with prominent intercellular spaces. The walls have many oval or rounded simple pits.

In one hypocotyl, in the lower transition region, a few strands of tertiary phloem were found in the xylem rays of the secondary external xylem. The strands were composed of only two, three, or four cells, as seen in cross section,

and their composition was not definitely established. The strands extended upward for a short distance and apparently ended blindly without branching.

Much less phloem than xylem is formed from the fascicular cambium. Even in an old root the secondary fascicular phloem may be no more than thirty cells wide, radially. There are numerous very large, rather short sieve tubes with small companion cells. Many parenchyma cells occur among the sieve tubes and in several definite phloem rays. In general, they are smaller than the sieve tubes, and may be practically isodiametric or somewhat elongated. They have many rounded simple pits.

As the amount of secondary thickening increases and the bundles become wider on the outer margin, the parenchyma of the primary phloem becomes active. The cell divisions and increased size of the cells increase the tangential and the radial diameters of the primary phloem so that it maintains, even in the old hypocotyls, its position over the entire outer face of the secondary phloem. In some cases it becomes a fairly thick layer of parenchyma with a few widely separated sieve tubes. The parenchyma cells are usually very short vertically and almost isodiametric or elongated tangentially.

The broad xylem rays over the protoxylem points are composed principally of parenchyma. The cells are almost isodiametric or elongated radially. There is a variable amount of tertiary phloem in the rays; in some rays, very little, in others there are many large phloem strands, particularly in

that part of the ray near the protoxylem point. These strands are similar to the tertiary strands in the pith. Most of them have a cambium zone on one side; some have a cambium extending partially around the strand; and some have no evident cambium. Some of the strands lie entirely in the ray. Others were initiated in the xylem parenchyma on the lateral face of the bundle and the ray parenchyma became involved later; or they were pushed out into the ray by the activity of a cambium between the strand and the xylem. In some places this lateral cambium extends for half the length of the face of the bundle and forms a wide band of phloem. There are smaller connecting phloem strands between these ray strands and the external interfascicular and fascicular phloem.

A few tertiary collateral bundles were found in the rays. Usually not more than one was found in a ray. The orientation of the bundles varies and the orientation of the same bundle may not be the same at different levels. Usually the phloem of the bundle faces the ray and, the xylem, the fascicular xylem. The course of the bundles has not been definitely established, but in some cases, though the phloem may continue for a long distance, the xylem extends only a short distance, ending blindly at its lower and upper ends. The structure of the bundles is similar to that of the tertiary bundles of the pith.

The external cambium and its derivatives in the transition region are similar in structure and activities to those in the root. In the upper transition region there are

numerous large horizontal phloem strands in the foliar rays connecting the tertiary phloem of the pith and the internal phloem with the external phloem. In the foliar rays there are a few tertiary phloem strands extending vertically.

Above the transition the secondary thickening is still essentially similar in structure to that of the root. The xylem and phloem rays are rather prominent. There is less tertiary phloem in the foliar rays, and that which is present is mainly in the form of strands connecting the external and internal phloems. Since the secondary bundles are not so wide on the outer face as in the root, the primary phloem group is not so large, though it is rather active. The group of primary phloem fibers is intact, even in very old hypocotyls.

The interfascicular cambium in some rays of the fairly old hypocotyl has formed a small amount of phloem, but most of the phloem in the foliar rays is tertiary.

The primary xylem of the bundles is almost always intact though occasionally some of the protoxylem elements have been crushed and resorbed.

The elements of the primary external interfascicular phloem are, in the old hypocotyls, widely spread apart by the activity of the parenchyma. The determination of the ultimate distribution of the primary phloem is made more difficult by the addition of tertiary phloem and by the secondary phloem internal to it.

3. The Internal Cambium and Its Derivatives

The internal cambium in the transition becomes active soon after the external cambium. Its activities have not been followed in detail, but they may be outlined in general. Usually the derivatives of the cambium are phloem cells toward the pith and parenchyma toward the primary xylem. Midway the transition this cambium may be an almost continuous ring. The phloem is similar to the external secondary phloem, though a much narrower band. In the oldest hypocotyls a small amount of xylem may be derived from this cambium, between it and the primary xylem. Thus a collateral bundle is formed with reverse orientation. These internal bundles are similar to the bundles of the pith. The phloem extends for a long distance up the axis, but the xylem, for only a short distance in some cases, ending blindly at its lower and upper ends.

Above the transition there is no internal cambium as a primary structure, but soon after a small amount of external secondary xylem has been formed from the external cambium, the parenchyma between the protoxylem and the internal phloem becomes active and forms a fairly definite cambium. This derived cambium cuts off parenchyma externally and phloem internally. Usually a larger amount of parenchyma than phloem is formed, though there are many more cells in the phloem. As a result the phloem strands in the oldest hypocotyls are found in the pith at a distance from the bundle. The parenchyma of the internal phloem, which lies just internal to the

secondary phloem, becomes active in the older hypocotyls, increasing slightly the thickness of the strand. In addition, there is in some, apparently, a small amount of tertiary phloem formed by the activity of the adjoining pith parenchyma. The primary phloem strands between the internal and the external phloem groups maintain their continuity even when there is a large amount of secondary thickening. These strands become larger by the addition of tertiary phloem and connect also with the internal secondary phloem.

The parenchyma derived from this internal cambium often becomes active, usually forming a few small strands of phloem near the primary xylem. In a few cases a small amount of xylem was formed, lying almost adjacent to the primary xylem and not closely associated with any phloem strand. The xylem consisted of one or two fairly large tracheae surrounded by small thick-walled cells with simple pits. It extended for a very short distance, ending blindly at its lower and upper ends.

4. The Pericycle

The pericycle in the root becomes active soon after secondary thickening has begun. The cell divisions are both periclinal and anticlinal, thus increasing tangentially and radially the size of these four rays of parenchymatous cells. Some of these cells toward the outer part mature rather early as phloem, though they are not derived from a definite cambium.

As the root increases in diameter and the cortex begins to disintegrate, the pericycle over the bundles becomes active, the cells dividing both periclinally and anticlinally, forming a layer three or four cells thick with a feeble periderm. This periderm is continuous over the broad xylem rays. At the same time the ray cells are dividing periclinally rather regularly in a line connecting the two adjacent fascicular cambiums, making a fairly continuous cambium ring.

As the root becomes older and larger the pericycle becomes more and more active, the cells dividing in many planes. In the oldest roots examined (three months) the pericycle forms a very irregular zone, about five or six cells thick, over the bundles, and somewhat thicker over the broad xylem rays. A fairly definite periderm is formed. Strands of phloem, irregularly arranged, are formed in the pericycle. They are more numerous over the broad xylem rays. These strands, usually composed of four or five cells, as seen in cross section, lie in all planes and form a very complex branching and anastomosing network with connections to the interfascicular secondary phloem.

The behavior of the pericycle in the transition is similar to that in the primary root. In the old hypocotyls there are usually more phloem strands than in the root. In the upper part of the transition and in the hypocotyl above this where the cortex persists, the pericycle is a narrow zone of actively dividing cells with a rather small number of strands of tertiary phloem extending in many directions. This band of

small cells extends disconnectedly over the foliar rays. There are connecting phloem strands to the interfascicular phloem in the foliar rays.

In the transition and upper hypocotyl, where the cortex has entirely disintegrated, a periderm is formed in the pericycle.

5. The Cortex

The cortex of the primary root dies and disintegrates fairly early. In a root only one millimeter in diameter with a small amount of secondary thickening the outer half of the cortex may be dead and crushed; and in a few places practically all of it may be gone. In a root about two millimeters in diameter all the cortex has disappeared. There seems to be a small amount of cell enlargement but practically no cell division in accomodation to the increased diameter of the root.

The cortex of the transition remains for a longer time, some still being present in a plant three weeks old with a moderate amount of secondary thickening. In the region of the peg a small portion of the cortex a few cells thick may be found in a hypocotyl three months old.

The cortex of the hypocotyl above the transition remains for a long time. It may be intact in a plant with a large amount of secondary thickening and from which the cortex of the transition region has been completely lost. Much of the inner region of the cortex may be present even in a hypocotyl

two months old and with a diameter of 15 mm. The cortical cells increase in tangential diameter and there are many cell divisions throughout, mostly anticlinal, but some periclinal. Above the upper part of the transition a definite, though somewhat irregular, periderm is usually formed in the inner region of the cortex a few cells external to the endodermis.

The outer cortical phloem is shed with the cortex, but the inner strands of the phloem may remain apparently unchanged in a hypocotyl three months old.

The endodermis remains on the axis only about as long as the rest of the cortex. In the root it is broken up by the increasing diameter of the stele, but no indications of cell divisions were found. In the upper part of the hypocotyl where the inner part of the cortex remains for a long time, the endodermis cannot be identified in the older hypocotyls. But since, external to many bundles, there is no layer of cells which have not divided, it is probable that the cells of the endodermis have divided in the older plants.

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